A CONTRIBUTION TO CYTOLOGY IN CORNALES[1]

PETER GOLDBLATT[2]

ABSTRACT

Chromosome counts in *Nyssa* and *Camptotheca* indicate a base number of $x = 22$ for Nyssaceae. *Davidia*, the only genus of Davidiaceae, has $x = 21$. In Cornaceae, where $x = 11$ was thought basic, *Mastixia* has $n = 13$ and 11, while *Curtisia*, recently excluded from Cornaceae as Curtisiaceae, has $n = 13$. Nyssaceae and Davidiaceae appear more closely related to Cornaceae than previously recognized subfamilies and tribes of this family, and segregation of *Helwingia*, *Mastixia* and *Curtisia* from Cornaceae as proposed by other workers is supported on cytological grounds.

While preparing a review of angiosperm cytology in relation to phylogeny, Raven (1975) was able to point out important gaps in the chromosome record for flowering plants. One of the groups where information was conspicuously lacking was the Cornaceae and its allied families Nyssaceae, Davidiaceae, and segregates such as Curtisiaceae.

An attempt was made to learn more of the cytology of these families, and by the time the review went to press (Raven, 1975), chromosome numbers had been determined for several rare and taxonomically critical genera. Most of these counts were available in time for inclusion in the review, but were not fully documented there. This paper, therefore, presents in detail the results of this recent chromosomal study with a short discussion of the possible significance to the systematics of the families concerned. The counts were made mainly by the writer but also by L. Rüdenberg and W. Tai, and with their permission, are also reported here.

MATERIALS, METHODS, AND OBSERVATIONS

Meiotic studies were made of most of the species investigated. Buds were fixed in Carnoy's solution (ethanol-acetic acid, 3:1) and stored in 70% ethanol.

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Counts for *Mastixia trichotoma* were made by W. Tai, Department of Botany and Plant Pathology, Michigan State University, while Lily Rüdenberg, Gray Herbarium, Harvard University, counted one of the collections of *Davidia involucrata* studied here.

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[2] B. A. Krukoff Curator of African Botany, Missouri Botanical Garden, 2345 Tower Grove Avenue, St. Louis, Missouri 63110, U.S.A.

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Mitotic studies were made in the case of *Mastixia trichotoma*, where a count was obtained from tapetal cells of developing anthers, and from root tips in *Camptotheca*, *Cornus capitata* and *Curtisia*. *Camptotheca* cuttings were supplied by the U.S. Department of Agriculture Plant Introduction Station, Chico, California, and grown at the Missouri Botanical Garden. *Curtisia* and *Cornus capitata* were grown from seed collected in the wild and also grown at the Missouri Botanical Garden. Root tips were pretreated in 0.003 M hydroxyquinoline for five hours at refrigerator temperature. After fixation they were hydrolyzed in 10% HCl and stained in lactopropionic orcein (Goldblatt, 1976). Chromosome counts obtained in the study are listed in Table 1, where collection data and voucher information is also given.

Discussion

At the time that this study was undertaken, the chromosome number was already well known in *Nyssa* with reports of $n = 22$ in *N. javanica* (Mehra & Bawa, 1969; Mehra, 1972) and in *N. sylvatica* (Dermen, 1932; Eyde, 1963; $n = 22 \pm 1$ being reported). The present study confirms $n = 22$ as fundamental in this genus, with reports for *N. ogeche* and *N. sylvatica* var. *biflora* representing first counts for these taxa.

In the other genus of Nyssaceae, the closely related *Camptotheca*, Raven (in Perdue, 1970) reported $n = 21$. Reexamining the same material of *C. acuminata* I find $n =$ ca. 22. Both records for *Camptotheca* were mitotic and in view of the numerous and small chromosomes in this species a meiotic count would be valuable in finally establishing its chromosome number.

In the monotypic *Davidia*, sometimes treated as a separate family, as here, following several recent authors (e.g., Cronquist, 1968), but otherwise placed in Nyssaceae, Dermen (1932) reported $n =$ ca. 20. The number for this close relative of Nyssaceae is now firmly established as $n = 21$ from two different sources, one count made by L. Rüdenberg and the other one by myself. This difference in chromosome number would support the belief that *Davidia* may be phylogenetically somewhat removed from *Nyssa* and *Camptotheca*, the two genera with which it is most often allied. The difference does not in itself necessarily imply familial separation, and the general relationship of *Davidia* to *Nyssa* and *Camptotheca* is not questioned. Fairbrothers (1977), on the basis of serological evidence also supports the separation of *Davidia* from *Nyssa* and *Camptotheca* and suggests subfamilial status for the two groups.

Cornaceae is a more heterogeneous family. As treated by Melchior (1964), following the classical treatment of Wangerin (1910), it was divided into three subfamilies, Curtisioideae, Mastixioideae, and Cornoideae, the latter with tribes Toricellieae, Helwingieae, Corneae, and Griselinieae. Treated here with Curtisioideae recognized as a separate family (Eyde, 1967; Takhtajan, 1970; Airy Shaw, 1972), Cornaceae remains the largest family in the cornalean alliance. A base number of $x = 11$ is well established for tribe Corneae, which also has $n = 10$, 9 in *Cornus* itself, with $n = 8$ in *Aucuba* and $n = 9$ in *Corokia*, both genera probably not correctly placed in Corneae (Adams, 1949) or even

TABLE 1. Chromosome counts for Cornaceae, Nyssaceae, Davidiaceae and Curtisiaceae obtained in this study. Voucher and collection information is presented as far as it is known.

Species	Chromosome No. *n*	Chromosome No. 2*n*	Collection and Voucher
	NYSSACEAE		
Nyssa sylvatica Marsh. var. *sylvatica*	22		Cultivated at U.S. National Arboretum (native of U.S.A.), *Eyde 120* (US).
N. sylvatica var. *biflora* (Walt.) Sargent	22		U.S.A., South Carolina, Berkeley Co., Santee swamp, *Priester s.n.* (MO).
N. ogeche Bartr. ex Marsh.	22		U.S.A., Florida, Liberty Co., *Godfrey 74223* (US).
Camptotheca acuminata Dec.		ca. 44	China, Chang An, Yung, Hsian (cult. U.S. Dept. Agriculture Plant Introduction Stn., Chico, California) (NA).
	DAVIDIACEAE		
Davidia involucrata Baill. var. *vilmoriniana* (Dode) Wangerin	21[a]		Cultivated at Arnold Arboretum, seed originally collected in Szech'uan, China, *Rehder s.n.* (AA).
	21		Cultivated at Strybing Arboretum, San Francisco, California, (CA).
	CORNACEAE—Mastixioideae		
Mastixia trichotoma Blume		22[b]	Indonesia, cult. at Cibodas Mountain Gardens, Bogor, (representative material at BO).
M. arborea (Wight) C. B. Cl.	13		India, Mysore State, Agumbe State Forest, Shimoge distr. *Matthew 14472* (US).
	CORNACEAE—Cornoideae		
Cornus capitata Wall.		22	Bhutan, E of Sintoka Dzong, *Bartholomew 209* (BM, UC).
	CURTISIACEAE		
Curtisia dentata (Burm. f.) C. A. Sm.		26	South Africa, Cape, Contour Path, Kirstenbosch, *Compton 8092* (NBG).

[a] Chromosome count made by Lily Rüdenberg, Gray Herbarium, Harvard University.
[b] Chromosome count made by W. Tai, Michigan State University.

Cornaceae (Eyde, 1964, 1967; Fairbrothers, 1977). The count of $n = 11$ reported here for *Cornus capitata* in fact confirms an earlier record for the species and clearly is representative of this tribe.

In the other tribes of Cornoideae, Griselinieae has as far as is known $x = 9$ ($n = 18$ in *Griselinia*, Federov, 1969; Raven, 1975) and Helwingeae $x = 19$ (Hara & Kurosawa, 1975). Helwingeae may in fact not be related at all to Cornaceae (see below). The remaining tribe, Toricellieae has $2n = 24$ (Malla et al., 1978), this based on a report for one of the three species of *Toricellia*, the only genus of the tribe. The count suggests that *Toricellia* might not be related to Cornaceae and the alternative placement in Araliaceae which has $x = 12$ (Raven, 1975) may be correct.

Counts here for *Mastixia*, the sole genus of Mastixioideae, are the first records for this subfamily. The situation in *Mastixia* with $n = 13$ and $n = 11$ in separate species is unclear as yet and appears complex. This range of chromosome number is unexpected in a genus of tropical, woody plants and requires confirmation. However, on present evidence $x = 13$ may be basic for Mastixioideae, which appears to support the placement of *Mastixia* in a separate subfamily of Cornaceae.

The monotypic African Curtisiaceae has a base number of $x = 13$, with $2n = 26$ in *Curtisia dentata*. If Curtisiaceae is, as generally accepted, fairly primitive in the cornalean alliance, the view that $x = 11$ is basic and fundamental in Cornales must be questioned. It seems that $x = 13$ or 14 may be ancestral with early reduction to 11 in Cornaceae and closely related families as Raven (1975: 740) postulated. The record of both $n = 13$ and 11 in Mastixioideae, though requiring further study, suggests a link between Curtisiaceae and Cornaceae.

With *Curtisia* and perhaps *Mastixia* excluded, $x = 11$ seems basic in Cornaceae. Evolution within Cornaceae appears to have involved aneuploid reduction, giving rise to $n = 10$ and 9 in Corneae and $x = 9$ in Griselinieae, assuming its position in Cornaceae is correct. *Helwingia*, with $n = 19$ firmly established, is problematic and its placement in Cornaceae has been seriously questioned by several systematists. Eyde (1966: 845; 1967: 177) supports its removal from Cornaceae on the basis of carpel vasculature not corresponding to the peculiar pattern in *Cornus* and its allies. Hutchinson (1967: 74) assigns *Helwingia* to Araliaceae where, however, its chromosome number conflicts sharply with $x = 12$ in all members of this family known cytologically. It is of interest to point out that *Helwingia*, of all Cornaceae and allied families, lacks the taxonomically significant iridoid compounds (Bate-Smith et al., 1975) and thus from a chemical point of view also seems best excluded from any cornalean alliance.

The two monogeneric families, Garryaceae and Alangiaceae, generally believed to be closely related to Cornaceae are known to have $x = 11$. The cytology certainly supports this view but does not exclude alternatives (Eyde, 1968, 1972).

Chromosome numbers appear to indicate a close relationship between Cornaceae-Cornoideae, Nyssaceae and Davidiaceae, with $x = 22$ in Nyssaceae possibly related by direct polyploidy to the $x = 11$, basic in Cornoideae. In fact, at least from a cytological point of view, Nyssaceae (and Davidiaceae) seem to be more closely related to Cornaceae than is Curtisiaceae. Recognition of this

family thus seems indicated on cytological as well as morphological and anatomical grounds. *Curtisia* also lacks the unusual carpel vasculature of Cornaceae, Nyssaceae, and Davidiaceae (Eyde, 1967).

There are still gaps in our knowledge of the cytology of Cornales and, in view of the information presented here, counts are most desirable for the two Madagascan genera, *Kaliphora* and *Melanophylla*, both of unknown affinities, though assigned by Wangerin (1910) to Cornaceae—Corneae and —Griselinieae, respectively. Cytological data on these two genera would be particularly valuable since both were excluded with *Curtisia* from Cornaceae by Eyde (1967) on anatomical grounds. *Melanophylla* has recently been placed in its own family (Takhtajan, 1970; Airy Shaw, 1972), while *Kaliphora* has been associated with the Afro-Madagascan Montiniaceae (Capuron, 1969; Krach, 1976; Dahlgren et al., 1977).

In summary, chromosome cytology can be said to suggest the following:

1. Nyssaceae, $n = 22$, and Davidiaceae, $n = 21$, stand out as being fundamentally hexaploid in the cornalean alliance and may be related by direct polyploidy to Cornaceae—Cornoideae, in which $x = 11$ is basic.

2. Cornaceae—Cornoideae—Corneae with $x = 11$ (and secondary aneuploidy) seem more closely related to the specialized Garryaceae and Alangiaceae (both also $x = 11$) than to Cornoideae—Griselinieae, —Toricellieae and —Helwingeae as well as to the traditionally recognized subfamilies of Cornaceae, Curtisioideae, and Mastixioideae.

3. Cytology provides support for the exclusion of *Curtisia* from Cornaceae based on other lines of evidence, while bringing into question the relationship of *Mastixia* and *Toricellia* to Cornaceae.

LITERATURE CITED

ADAMS, J. E. 1949. Studies in the comparative anatomy of the Cornaceae. J. Elisha Mitchell Sci. Soc. 65: 218–244.

AIRY SHAW, H. K. 1972. A new species of *Melanophylla* Baker (Melanophyllaceae). Kew Bull. 26:491–493.

BATE-SMITH, E. C., I. K. FERGUSON, K. HUTSON, S. R. JENSEN, B. J. NIELSEN & T. SWAIN. 1975. Phytochemical interrelationships in the Cornaceae. Biochem. Syst. Ecol. 3: 79–89.

CAPURON, R. 1969. Contribution à l'étude de la flore forestière de Madagascar. Sur la place du genre *Kaliphora* Hook. f. Adansonia, sér. 2, 9: 395–397.

CRONQUIST, A. 1968. The Evolution and Classification of Flowering Plants. Houghton Mifflin, Boston.

DAHLGREN, R., S. ROSENDAL & B. J. NIELSEN. 1977. Seedling morphology and iridoid occurrence in *Montinia caryophyllacea* (Montiniaceae). Bot. Not. 130: 329–332.

DERMEN, H. 1932. Cytological studies of *Cornus*. J. Arnold Arbor. 13: 410–415.

EYDE, R. H. 1963. Morphological and paleobotanical studies of the Nyssaceae, I. A survey of the modern species and their fruit. J. Arnold Arbor. 44: 1–52.

———. 1964. Inferior ovary and generic affinities of *Garrya*. Amer. J. Bot. 51: 1083–1092.

———. 1966. Systematic anatomy of the flower and fruit of *Corokia*. Amer. J. Bot. 53: 833–847.

———. 1967. The peculiar gynoecial vasculature of Cornaceae and its systematic significance. Phytomorphology 17: 172–182.

———. 1968. Flowers, fruits and phylogeny of Alangiaceae. J. Arnold Arbor. 49: 167–192.

———. 1972. Pollen of *Alangium*: toward a more satisfactory synthesis. Taxon 21: 471–477.

FAIRBROTHERS, D. E. 1977. Perspectives in plant serotaxonomy. Ann. Missouri Bot. Gard. 64: 147–160.
FEDEROV, A. 1969. Chromosome Numbers of Flowering Plants. Scientific Press, Leningrad.
GOLDBLATT, P. 1976. Cytotaxonomic studies in the tribe Quillajeae (Rosaceae). Ann. Missouri Bot. Gard. 62: 200–206.
HARA, H. & S. KUROSAWA. 1975. A revision of the genus *Helwingia*. Pp. 393–413, *in* H. Ohashi (editor), The Flora of Eastern Himalaya. Third Report. Univ. of Tokyo Press, Tokyo.
HUTCHINSON, J. 1967. The Genera of Flowering Plants. Vol. 2. Clarendon Press, Oxford.
KRACH, J. E. 1976. Die Samen der Saxifragaceae. Bot. Jahrb. Syst. 97: 1–60.
MALLA, S. B., S. BHATTARAI, M. GORKHALI, H. SAIJU & M. KAYASTHA. 1978. *In* IOPB chromosome number reports LIX. Taxon 27: 53–61.
MEHRA, P. N. 1972. Cytogenetical evolution of hardwoods. Nucleus 15: 64–83.
——— & K. S. BAWA. 1969. Chromosomal evolution in tropical hardwoods. Evolution 23: 466–481.
MELCHIOR, E. (editor). 1964. Engler's Syllabus der Pfanzenfamilien. Vol. 2. Borntraeger, Berlin.
PERDUE, R. E. 1970. *Camptotheca acuminata* Decaisne (Nyssaceae). Source of camptothecin, an antileukemic alkaloid. Techn. Bull. U.S.D.A. 1415: 1–26.
RAVEN, P. H. 1975. The bases of Angiosperm phylogeny: Cytology. Ann. Missouri Bot. Gard. 62: 724–764.
TAKHTAJAN, A. 1970. Origin and Dispersal of the Flowering Plants. Scientific Press, Leningrad. [In Russian.]
WANGERIN, W. 1910. Garryaceae, Nyssaceae, Alangiaceae, Cornaceae. Das Pflanzenreich IV. 56a, 220a, 220b, 229 (Heft 41): 1–17, 1–19, 1–24, 1–110. Englemann, Leipzig.